

## SEQUENCE LISTING

&lt;110&gt; EBL GmbH

&lt;120&gt; Method for the production of protamine

&lt;130&gt; Protamin

&lt;140&gt;

&lt;141&gt;

&lt;160&gt; 36

&lt;170&gt; PatentIn Ver. 2.1

&lt;210&gt; 1

&lt;211&gt; 102

&lt;212&gt; DNA

&lt;213&gt; Oncorhynchus mykiss

&lt;220&gt;

&lt;221&gt; CDS

&lt;222&gt; (1)..(99)

&lt;220&gt;

<223> aa sequence derived from ORF of nucleotide  
sequence

&lt;400&gt; 1

atg	ccc	aga	aga	cgc	aga	tcc	tcc	agc	cga	cct	gtc	cgc	agg	cgc	cgc	48
Met	Pro	Arg	Arg	Arg	Arg	Ser	Ser	Ser	Arg	Pro	Val	Arg	Arg	Arg	Arg	
1				5				10				15				
cgc	ccc	agg	gtg	tcc	cga	cgt	cgt	cgc	agg	aga	gga	ggc	cgc	agg	agg	96
Arg	Pro	Arg	Val	Ser	Arg	Arg	Arg	Arg	Arg	Arg	Gly	Gly	Arg	Arg	Arg	
			20					25				30				
cgt	tag															102
Arg																

&lt;210&gt; 2

&lt;211&gt; 33

&lt;212&gt; PRT

&lt;213&gt; Oncorhynchus mykiss

<223> aa sequence derived from ORF of nucleotide  
sequence

&lt;400&gt; 2

Met Pro Arg Arg Arg Arg Ser Ser Ser Arg Pro Val Arg Arg Arg Arg  
 1 5 10 15

Arg Pro Arg Val Ser Arg Arg Arg Arg Arg Arg Gly Gly Arg Arg Arg  
 20 25 30

Arg

&lt;210&gt; 3

&lt;211&gt; 102

&lt;212&gt; DNA

&lt;213&gt; Oncorhynchus mykiss

&lt;220&gt;

&lt;221&gt; CDS

&lt;222&gt; (1)..(99)

&lt;220&gt;

<223> aa sequence derived from ORF of nucleotide  
 sequence

&lt;400&gt; 3

atg ccc aga aga cgc aga tcc tcc aga cca cct gtc cgc agg cgc cgc 48  
 Met Pro Arg Arg Arg Arg Ser Ser Arg Pro Pro Val Arg Arg Arg Arg  
 1 5 10 15

cgc ccc agg gtg tcc cga cgt cgt cgc agg aga gga ggc cgc agg agg 96  
 Arg Pro Arg Val Ser Arg Arg Arg Arg Arg Arg Arg Gly Gly Arg Arg Arg  
 20 25 30

cgt tag

102

Arg

&lt;210&gt; 4

&lt;211&gt; 33

&lt;212&gt; PRT

&lt;213&gt; Oncorhynchus mykiss

<223> aa sequence derived from ORF of nucleotide  
 sequence

&lt;400&gt; 4

Met Pro Arg Arg Arg Arg Ser Ser Arg Pro Pro Val Arg Arg Arg Arg  
 1 5 10 15

Arg Pro Arg Val Ser Arg Arg Arg Arg Arg Arg Gly Gly Arg Arg Arg  
                   20                                  25                                  30

Arg

<210> 5

<211> 102

<212> DNA

<213> Oncorhynchus mykiss

<220>

<221> CDS

<222> (1)..(99)

<220>

<223> aa sequence derived from ORF of nucleotide  
                   sequence

<400> 5

atg ccc aga aga cgc aga tcc tcc aga cga cct gtc cgc agg cgc cgc 48  
 Met Pro Arg Arg Arg Arg Ser Ser Arg Arg Pro Val Arg Arg Arg Arg  
       1                                  5                                  10                                  15

cgc ccc agg gtg tcc cga cgt cgt cgc agg aga gga ggc cgc agg agg 96  
 Arg Pro Arg Val Ser Arg Arg Arg Arg Arg Arg Gly Gly Arg Arg Arg  
                   20                                  25                                  30

cgt tag 102  
 Arg

<210> 6

<211> 33

<212> PRT

<213> Oncorhynchus mykiss

<223> aa sequence derived from ORF of nucleotide  
                   sequence

<400> 6

Met Pro Arg Arg Arg Arg Ser Ser Arg Arg Pro Val Arg Arg Arg Arg  
       1                                  5                                  10                                  15

Arg Pro Arg Val Ser Arg Arg Arg Arg Arg Arg Gly Gly Arg Arg Arg  
                   20                                  25                                  30

Arg

&lt;210&gt; 7

&lt;211&gt; 102

&lt;212&gt; DNA

&lt;213&gt; Oncorhynchus mykiss

&lt;220&gt;

&lt;221&gt; CDS

&lt;222&gt; (1)..(99)

&lt;220&gt;

<223> aa sequence derived from ORF of nucleotide  
sequence

&lt;400&gt; 7

atg	ccc	aga	aga	cgc	aga	tcc	tct	agc	cga	cct	gtc	cgc	agg	cgc	cgc	48
Met	Pro	Arg	Arg	Arg	Arg	Ser	Ser	Ser	Arg	Pro	Val	Arg	Arg	Arg	Arg	
1				5					10					15		

cgc	gcc	agg	gtg	tcc	cga	cgt	cgt	cgc	agg	aga	gga	cgc	cgc	agg	agg	96
Arg	Ala	Arg	Val	Ser	Arg	Arg	Arg	Arg	Arg	Arg	Gly	Arg	Arg	Arg	Arg	
			20					25					30			

cgt	tag															102
Arg																

&lt;210&gt; 8

&lt;211&gt; 33

&lt;212&gt; PRT

&lt;213&gt; Oncorhynchus mykiss

<223> aa sequence derived from ORF of nucleotide  
sequence

&lt;400&gt; 8

Met	Pro	Arg	Arg	Arg	Arg	Ser	Ser	Ser	Arg	Pro	Val	Arg	Arg	Arg	Arg
1					5				10					15	

Arg	Ala	Arg	Val	Ser	Arg	Arg	Arg	Arg	Arg	Arg	Gly	Arg	Arg	Arg	Arg
			20					25						30	

Arg

<210> 9  
 <211> 102  
 <212> DNA  
 <213> Oncorhynchus mykiss

<220>  
 <221> CDS  
 <222> (1)..(99)

<220>  
 <223> aa sequence derived from ORF of nucleotide  
 sequence

<400> 9  
 atg ccc aga aga cgc aga tcc tcc agc cga cct gtc cgc agg cgc cgc 48  
 Met Pro Arg Arg Arg Arg Ser Ser Ser Arg Pro Val Arg Arg Arg Arg  
 1 5 10 15  
 cgc ccc agg gtg tcc cga cgt cgt cgc agg aga gga cgc cgc agg agg 96  
 Arg Pro Arg Val Ser Arg Arg Arg Arg Arg Arg Gly Arg Arg Arg Arg  
 20 25 30  
 cgt tag 102  
 Arg

<210> 10  
 <211> 33  
 <212> PRT  
 <213> Oncorhynchus mykiss  
 <223> aa sequence derived from ORF of nucleotide  
 sequence

<400> 10  
 Met Pro Arg Arg Arg Arg Ser Ser Ser Arg Pro Val Arg Arg Arg Arg  
 1 5 10 15  
 Arg Pro Arg Val Ser Arg Arg Arg Arg Arg Arg Gly Arg Arg Arg Arg  
 20 25 30  
 Arg

<210> 11  
 <211> 102  
 <212> DNA  
 <213> Oncorhynchus keta

<220>  
 <221> CDS  
 <222> (1)..(99)

<220>  
 <223> aa sequence derived from ORF of nucleotide  
 sequence

<400> 11  
 atg ccc aga aga cgc aga tcc tcc agc cga cct gtc cgc agg cgc cgc 48  
 Met Pro Arg Arg Arg Arg Ser Ser Ser Arg Pro Val Arg Arg Arg Arg  
 1 5 10 15  
 cgc cct agg gtg tcc cga cgt cgt cgc agg aga gga ggc cgc agg agg 96  
 Arg Pro Arg Val Ser Arg Arg Arg Arg Arg Arg Gly Gly Arg Arg Arg  
 20 25 30  
 cgt tag 102  
 Arg

<210> 12  
 <211> 33  
 <212> PRT  
 <213> Oncorhynchus keta  
 <223> aa sequence derived from ORF of nucleotide  
 sequence

<400> 12  
 Met Pro Arg Arg Arg Arg Ser Ser Ser Arg Pro Val Arg Arg Arg Arg  
 1 5 10 15  
 Arg Pro Arg Val Ser Arg Arg Arg Arg Arg Arg Gly Gly Arg Arg Arg  
 20 25 30  
 Arg

<210> 13  
 <211> 102  
 <212> DNA

<213> Oncorhynchus mykiss

<220>

<221> CDS

<222> (1)..(99)

<220>

<223> nucleotide sequence derived from amino acid  
sequence

<400> 13

atg ccc aga aga cgc aga tcc tcc agc cga cct gtc cgc agg cgc cgc 48  
Met Pro Arg Arg Arg Arg Ser Ser Ser Arg Pro Val Arg Arg Arg Arg  
1 5 10 15

cgc gcn agg gtg tcc cga cgt cgt cgc agg aga gga ggc cgc agg agg 96  
Arg Ala Arg Val Ser Arg Arg Arg Arg Arg Arg Gly Gly Arg Arg Arg  
20 25 30

cgt tag 102  
Arg

<210> 14

<211> 33

<212> PRT

<213> Oncorhynchus mykiss

<223> nucleotide sequence derived from amino acid  
sequence

<400> 14

Met Pro Arg Arg Arg Arg Ser Ser Ser Arg Pro Val Arg Arg Arg Arg  
1 5 10 15

Arg Ala Arg Val Ser Arg Arg Arg Arg Arg Arg Gly Gly Arg Arg Arg  
20 25 30

Arg

<210> 15

<211> 96

<212> DNA

<213> Oncorhynchus mykiss

<220>

&lt;221&gt; CDS

&lt;222&gt; (1)..(93)

&lt;220&gt;

<223> nucleotide sequence derived from amino acid  
sequence

&lt;400&gt; 15

atg	ccc	aga	aga	cgc	aga	gcn	agc	cga	cgn	gtc	cgc	agg	cgc	cgc	cgc	48
Met	Pro	Arg	Arg	Arg	Arg	Ala	Ser	Arg	Arg	Val	Arg	Arg	Arg	Arg	Arg	
1				5						10				15		

ccc	agg	gtg	tcc	cga	cgt	cgc	agg	aga	gga	ggc	cgc	agg	agg	cgt	tag	96
Pro	Arg	Val	Ser	Arg	Arg	Arg	Arg	Arg	Gly	Gly	Arg	Arg	Arg	Arg	Arg	
			20					25					30			

&lt;210&gt; 16

&lt;211&gt; 31

&lt;212&gt; PRT

&lt;213&gt; Oncorhynchus mykiss

<223> nucleotide sequence derived from amino acid  
sequence

&lt;400&gt; 16

Met	Pro	Arg	Arg	Arg	Arg	Ala	Ser	Arg	Arg	Val	Arg	Arg	Arg	Arg	Arg	
1				5						10				15		

Pro	Arg	Val	Ser	Arg	Arg	Arg	Arg	Arg	Gly	Gly	Arg	Arg	Arg	Arg	Arg	
			20					25					30			

&lt;210&gt; 17

&lt;211&gt; 96

&lt;212&gt; DNA

&lt;213&gt; Oncorhynchus mykiss

&lt;220&gt;

&lt;221&gt; CDS

&lt;222&gt; (1)..(93)

&lt;220&gt;

<223> nucleotide sequence derived from amino acid  
sequence

&lt;400&gt; 17

atg	ccc	aga	aga	cgc	aga	gcn	agc	cga	cgn	ath	cgc	agg	cgc	cgc	cgc	48
-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	----



Met Pro Arg Arg Arg Arg Ala Ser Arg Arg Ile Arg Arg Arg Arg Arg  
 1 5 10 15

ccc agg gtg tcc cga cgt cgc agg aga gga ggc cgc agg agg cgt tag 96  
 Pro Arg Val Ser Arg Arg Arg Arg Arg Gly Gly Arg Arg Arg Arg  
 20 25 30

<210> 18

<211> 31

<212> PRT

<213> Oncorhynchus mykiss

<223> nucleotide sequence derived from amino acid  
 sequence

<400> 18

Met Pro Arg Arg Arg Arg Ala Ser Arg Arg Ile Arg Arg Arg Arg Arg  
 1 5 10 15

Pro Arg Val Ser Arg Arg Arg Arg Arg Gly Gly Arg Arg Arg Arg  
 20 25 30

<210> 19

<211> 102

<212> DNA

<213> Oncorhynchus mykiss

<220>

<221> CDS

<222> (1)..(99)

<220>

<223> nucleotide sequence derived from amino acid  
 sequence

<400> 19

atg ccc agā agā cgc aga aga tcc tcc agc cga cct ath cgc agg cgc - 48 -  
 Met Pro Arg Arg Arg Arg Ser Ser Ser Arg Pro Ile Arg Arg Arg  
 1 5 10 15

cgc cgc ccc agg gtg tcc cga cgt cgc agg aga gga ggc cgc agg agg 96  
 Arg Arg Pro Arg Val Ser Arg Arg Arg Arg Arg Gly Gly Arg Arg Arg  
 20 25 30

cgt tag

Arg

102

<210> 20  
 <211> 33  
 <212> PRT  
 <213> Oncorhynchus mykiss  
 <223> nucleotide sequence derived from amino acid  
 sequence

<400> 20  
 Met Pro Arg Arg Arg Arg Arg Ser Ser Ser Arg Pro Ile Arg Arg Arg  
           1                  5                  10                  15  
 Arg Arg Pro Arg Val Ser Arg Arg Arg Arg Arg Gly Gly Arg Arg Arg  
                   20                  25                  30  
 Arg

<210> 21  
 <211> 96  
 <212> DNA  
 <213> Clupea harengus

<220>  
 <221> CDS  
 <222> (1)..(93)  
 <220>  
 <223> nucleotide sequence derived from amino acid  
 sequence

<400> 21  
 atg ccc aga aga cgc acc aga cgc gcn agc cga cct gtc cgc agg cgc 48  
 Met Pro Arg Arg Arg Thr Arg Arg Ala Ser Arg Pro Val Arg Arg Arg  
           1                  5                  10                  15  
 cgc ccc agg cgc gtg tcc cga cgt cgt cgc gca cgc cgc agg agg tag 96  
 Arg Pro Arg Arg Val Ser Arg Arg Arg Arg Ala Arg Arg Arg Arg Arg  
                   20                  25                  30

<210> 22  
 <211> 31  
 <212> PRT  
 <213> Clupea harengus

<223> nucleotide sequence derived from amino acid  
sequence

<400> 22

Met Pro Arg Arg Arg Thr Arg Arg Ala Ser Arg Pro Val Arg Arg Arg  
1 5 10 15

Arg Pro Arg Arg Val Ser Arg Arg Arg Arg Ala Arg Arg Arg Arg  
20 25 30

<210> 23

<211> 99

<212> DNA

<213> Clupea harengus

<220>

<221> CDS

<222> (1)..(96)

<220>

<223> nucleotide sequence derived from amino acid  
sequence

<400> 23

atg gcc aga aga cgc aga agc aga cgc gcn agc cga cct gtc cgc agg 48  
Met Ala Arg Arg Arg Ser Arg Arg Ala Ser Arg Pro Val Arg Arg  
1 5 10 15

cgc cgc ccc agg cgc gtg tcc cga cgt cgt cgc gca cgc cgc agg agg 96  
Arg Arg Pro Arg Arg Val Ser Arg Arg Arg Arg Ala Arg Arg Arg Arg  
20 25 30

tag 99

<210> 24

<211> 32

<212> PRT

<213> Clupea harengus

<223> nucleotide sequence derived from amino acid  
sequence

<400> 24

Met Ala Arg Arg Arg Arg Ser Arg Arg Ala Ser Arg Pro Val Arg Arg  
1 5 10 15

Arg Arg Pro Arg Arg Val Ser Arg Arg Arg Arg Ala Arg Arg Arg Arg  
20 25 30

<210> 25

<211> 99

<212> DNA

<213> Clupea harengus

<220>

<221> CDS

<222> (1) .. (96)

**<220>**

<223> nucleotide sequence derived from amino acid  
sequence

<400> 25

atg gcc aga aga cgc aga tcc tcc agc cga cct ath cgc agg cgc cgc 48  
Met Ala Arg Arg Arg Arg Ser Ser Ser Arg Pro Ile Arg Arg Arg Arg  
1 5 10 15

ccc agg cgc cgg acc aca cgt cgt cgc agg gca ggc cgc agg agg cgt 96  
Pro Arg Arg Arg Thr Thr Arg Arg Arg Arg Ala Gly Arg Arg Arg Arg  
20 25 30

tag 99

<210> 26

<211> 32

<212> PRT

<213> Clupea harengus

<223> nucleotide sequence derived from amino acid  
sequence

<400> 26

Met-Ala-Arg Arg Arg Arg-Ser Ser Ser Arg-Pro Ile Arg Arg-Arg-Arg -  
1 5 10 15

Pro Arg Arg Arg Thr Thr Arg Arg Arg Arg Ala Gly Arg Arg Arg Arg  
20 25 30

<210> 27

$\langle 211 \rangle$  111

<212> DNA  
<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: consensus 1

<400> 27

atgscagaa gacgcagaas cagaysckn agmcsacstr thcgcaggcg ccgccgcscy 60  
aggcgcskgw ccmsacgtcg tcgcaggaga gsasgccgca ggaggcgta g 111

<210> 28

<211> 102

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: consensus 2

<400> 28

atgccccgnc gncgccgntc ctccagccga cctgtccgcc gncgccgccg cccccngtg 60  
tcccgacgtc gtcgccgncg nggaggccgc cgnccgncgtt ag 102

<210> 29

<211> 102

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: consensus 3

<400> 29

atgccgcggc gccgccggtc gtcgagccgc ccggtgcgtc gccggcgccg cccgcgggtc 60  
tcgcgccgcc gccggcgccg cggcggccgc cggcgccgct ga 102

<210> 30

<211> 102

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: consensus 4

<400> 30

atgccgcgcc gtcgccgtag ctcgagccgt ccggtgcgtc gccgtcgccg tccccgtgtc 60  
agccgccgcc gccgtcgccg cggcggaacgc cgtcgccgtt ga 102

<210> 31  
<211> 102  
<212> DNA  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: consensus 5

<400> 31  
atgccgcggc gtcggcgag ctccagccgt ccagtgcggc gccgtcgccg ccccggtgtc 60  
tcgcgccgcc gccggcgccg cggcggacgc cgtcgccggt ga 102

<210> 32  
<211> 102  
<212> DNA  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: ebl 1

<400> 32  
atgcgcggc gtcggcgtag ctccagccgt ccagtgcgtc gccgtcgccg ccccggtgtc 60  
tcgcgccgcc gccggcgccg cggcggacgc cgtcgccggt ga 102

<210> 33  
<211> 36  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> aa position 1: X= zero or M

<220>  
<223> aa position 2: X= A or P

<220>  
<223> aa position 6: X= zero or R

<220>  
<223> aa position 7: X=zero or T or S

<220>  
<223> aa position 8: X= zero or R

<220>

<223> aa position 9: X=zero or R or S

<220>

<223> aa position 10: X= S or A

<220>

<223> aa position 11: X= S or R

<220>

<223> aa position 12: X= R or P

<220>

<223> aa position 13: X= P or R

<220>

<223> aa position 14: X= V or I

<220>

<223> aa position 19: X= zero or R

<220>

<223> aa position 20: X= P or A

<220>

<223> aa position 22: X= zero or R

<220>

<223> aa position 23: X= V or R

<220>

<223> aa position 24: X= S or T

<220>

<223> aa position 25: X= R or T

<220>

<223> aa position 29: X= zero or R

<220>

<223> aa position 30: X= zero or R

<220>

<223> aa position 31: X= G or A

<220>

<223> aa position 32: X= G or R

<220>

<223> aa position 36: X= zero or R

<220>

<223> Description of Artificial Sequence: consensus  
sequence

<400> 33

Xaa Xaa Arg Arg Arg Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Arg Arg  
1 5 10 15

Arg Arg Xaa Xaa Arg Xaa Xaa Xaa Xaa Arg Arg Arg Xaa Xaa Xaa Xaa  
20 25 30

Arg Arg Arg Xaa  
35

<210> 34

<211> 227

<212> DNA

<213> Artificial Sequence

<220>

<221> CDS

<222> (43)..(108)

<220>

<221> CDS

<222> (109)..(207)

<220>

<223> Description of Artificial Sequence: cloning  
sequence for expression of Protamine

<220>

<221> sig\_peptide

<222> (43)..(108)

<223> pelB gene

<220>

<221> misc\_feature

<222> (1)..(6)

<223> XbaI restriction site

<220>

<221> misc\_feature

<222> (222)..(227)



<223> Bam HI restriction site

<220>

<221> RBS

<222> (28)..(33)

<223> IRES sequence

<220>

<221> gene

<222> (109)..(207)

<223> ebl 1 gene

<400> 34

tctagaaata attttgttta actttaagaa ggagatatac at atg aaa tac ctg 54  
Met Lys Tyr Leu  
1

ctg ccg acc gct gct gct ggt ctg ctg ctc ctc gct gcc cag ccg gcg 102  
Leu Pro Thr Ala Ala Ala Gly Leu Leu Leu Leu Ala Ala Gln Pro Ala  
5 10 15 20

atg gcc atg ccg cgg cgt cgg cgt agc tcc agc cgt cca gtg cgt cgc 150  
Met Ala Met Pro Arg Arg Arg Arg Ser Ser Ser Arg Pro Val Arg Arg  
25 30 35

cgt cgc cgc ccc cgt gtc tcg cgc cgc cgc cgg cgc cgc ggc gga cgc 198  
Arg Arg Arg Pro Arg Val Ser Arg Arg Arg Arg Arg Arg Gly Gly Arg  
40 45 50

cgt cgc cgt tgaggaatta attcggatcc 227  
Arg Arg Arg  
55

<210> 35

<211> 22

<212> PRT

<213> Artificial Sequence

<223> Description of Artificial Sequence: cloning  
sequence for expression of Protamine

<400> 35

Met Lys Tyr Leu Leu Pro Thr Ala Ala Ala Gly Leu Leu Leu Ala  
1 5 10 15  
Ala Gln Pro Ala Met Ala  
20

<210> 36

<211> 33

<212> PRT

<213> Artificial Sequence

<223> Description of Artificial Sequence: cloning  
sequence for expression of Protamine

<400> 36

Met Pro Arg Arg Arg Arg Ser Ser Ser Arg Pro Val Arg Arg Arg Arg

1

5

10

15

Arg Pro Arg Val Ser Arg Arg Arg Arg Arg Arg Gly Gly Arg Arg Arg

20

25

30

Arg